Effects of Haul-out Bucket Plunge Height and Water-to-Water Transfer of Threadfin Shad and Delta Smelt at the Tracy Fish Collection Facility

Investigators

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Summary

Fish salvage operations of the Tracy Fish Collection Facility (TFCF) at the head of the Delta Mendota Canal in California require daily collection and holding of fish, and the transport of these fish back to the Sacramento-San Joaquin River Delta, away from the facilities. These functions are of major importance for efficient return and survival of salvaged fishes; however collecting, handling, and transport methods associated with entrainment of the fishes inadvertently may cause harm that the fish salvage facilities are attempting to minimize. The Bureau of Reclamation (Reclamation) Tracy Fish Collection Facility consists of a system of louvers, bypasses, collecting/holding tanks, transfer haul-out buckets, and trucking to reduce the associated fish loss of its pumping operation. Evaluations and improvements of both the state and federal fish salvage facilities have been ongoing for a number of years. There has been continued focus on the affects of fish transfer from collecting/holding tank to transport truck and this is thought to be one of the most harmful components of the fish salvage process. Recently a new fish haul-out bucket (1,544 L) was designed for water-to-water fish transfer and put into operation. It has two main improvements over the old bucket. First the drain hole was enlarged from 10–12 inches to reduce debris from clogging the drain hole. Second, the bucket was designed for water-to-water fish transfer into the fish truck. The water-to-water transfer method takes more time to complete and its effectiveness in increasing 20-mm larval and adult fish. Regardless of the new transfer mechanism, the operation of the bucket is still suspect and needs continued evaluation. We suggest an assessment of the current manner at which the haul-out bucket is operated compared to that of a water-to-water procedure and with a rubber gasket mounted to the filling hole of the transport truck.

Direct free fall of water from the haul-out bucket into the truck tank may cause sheer force stress on fish. In addition, fish are in debris that may accumulate near the hole. Water volume pressure pushing salvaged fish through a narrow drain may cause stress and inflict bodily harm, potentially leading to death. Measuring the acute physiological stress and potential direct and indirect mortality experienced by fishes during the transferring of fish in the haul-out bucket to the fish-haul truck is vital to understanding negative impacts the process may have on fish. Evaluating physiological responses of fish to plunging depths and haul-out bucket release stressors can be measured using blood plasma constituents such as cortisol, lactate, and glucose. The degree of bodily damage and scaled loss can be determined using a fluorescein dye procedure (Noga and Udomkusonsri 2002). Finally, and most important, fish should be held for 96 h after the potential stressor to determine stressor-associated deaths. Information will be used to identify and evaluate potential impacts of using the haul-out bucket and provide recommendations on a standard operating procedure on the best way to operate this transfer method.

Problem Statement

Fish transfer mechanisms (*i.e.*, haul-out bucket) and their operations are a major concern to the safe and effective release of salvaged fishes. There is evidence that this vital link in the process may be one of the most harmful components of the fish salvage process (Portz 2007). Recently a new 1,544.3-L fish haul-out bucket was implemented to reduce harmful effects of salvaging fish. However, the operation of the bucket is still suspected of impacting fish, and the operating procedures need continued evaluation. We suggest a laboratory assessment of the current manner at which the haul-out bucket is operated compared to that of a water-to-water procedure and with a rubber gasket mounted to the filling hole of the transport truck using the simulated haul-out-bucket in the Denver Hydraulics Laboratory.

Goals and Hypotheses

Goals:

- 1. Determine if threadfin shad and larval/juvenile delta smelt are physiologically compromised by the pressure and free fall from haul-out bucket evacuation into the fish hauling truck.
- 2. Determine if pressures exerted when emptying the haul-out bucket and free fall into the fish hauling truck affect scale loss and/or external tissue damage in threadfin shad and larval/juvenile delta smelt.
- 3. Determine if pressure and free fall from haul-out bucket evacuation affects the short-term survival (96 h) of threadfin shad and larval/juvenile delta smelt.
- 4. Provide evidence and recommendations that will support a more effective standard operating procedure for haul-out bucket use for safely transferring salvaged fish from collecting/holding tanks to transport truck. The use of water-to-water transferring procedures and using a rubber gasket mounted to the filling hole of the transport truck should be investigated.

Hypotheses:

- 1. If pressure and free fall experienced by threadfin shad and larval/juvenile delta smelt when emptying the haul-out bucket are physiological stressful, then heightened plasma cortisol, glucose, and lactate concentrations should result compared to water-to-water transferring procedures.
- 2. If pressure and free fall experienced by threadfin shad and larval/juvenile delta smelt when emptying the haul-out bucket affects scale loss and external tissue damage, then fish experiencing this transfer technique will have greater areas of skin ulcerations and damage compared to control fish and fish undergoing water-to-water transferring procedures.
- 3. If a pressure and free fall experienced by threadfin shad and larval/juvenile delta smelt when emptying the haul-out bucket affects the short-term survival (96 h), then fish experiencing this transfer technique will have greater mortality compared to control fish and fish undergoing water-to-water transferring procedures.

Materials and Methods

Source and Care of Fish

Threadfin shad (*Dorosoma petenense*) will be obtained from a commercial fish farm and larval/juvenile delta smelt (*Hypomesus transpacificus*) from University of California Davis' Fish Conservation and Culture Lab in Byron, California) and transported to the Denver Federal Center (Denver, Colorado). Threadfin shad and delta smelt will be maintained in 757-L circular tanks equipped with aerated, recirculated dechlorinated municipal water. Fish will be held under a natural photoperiod (39° 43' N latitude) with natural and halogen light, and fed at 2% body weight per day. Treatment and control fish may be marked with implanted, colored microspheres on dorsal and anal fins with a high pressure needle (Photonic tagging; New West Technology, Arcata, California) or calcein batch marking to differentiate various treatment fish so they can be consolidated for a 96-h survival holding period.

The Experiment: Effects Of Plunge Height and Water-to-Water Transfer on Physiological Stress, Body Damage, and Survival.

The experiment will be organized to evaluate the physiological stress response, scale loss and external tissue damage, and short-term survival (96 h) of threadfin shad and larval/juvenile delta smelt experiencing free fall from the haul-out bucket along with water volume pressure pushing salvaged fish through its narrow drain. Fifty treatment fish will be placed in a simulated haul-out bucket and emptied into an external 757-L tank from an operational plunge height that is currently being used at the TFCF. Additionally, a water-to-water transferring method will be tested where there is no plunge depth or free fall, and fish are evacuated from the bucket without air exposure. Water quality (*i.e.*, temperature, dissolved oxygen concentration) will be monitored throughout the study along with a detailed description of physical parameters (*i.e.*, fall height, diameters, and pressures). A minimum of 36 replicates for each species will be collected testing haul-out bucket procedures.

Physiological Stress Response

A control fish will be captured and removed from previously undisturbed tanks with modified 10- × 18-cm dip nets with a 1.5-L plastic reservoir sewn into the cod-end, so that fish could be transferred in water to minimize stress. All transfers of control fish will be accomplished quickly (<30 s) with minimal disturbance and handling trauma to the fish. Fifty treatment fish will be nettled, placed in an 18.9-L bucket, and inserted in the simulated haul-out bucket. Treatment fish after recapture will be quickly transferred to a bath containing a lethal dose of tricaine methanesulfate (MS-222, Argent Chemical Laboratories, Inc., Redmond, Washington; 200 mg/L), which immobilizes them in less than 30 s. This anesthetic dose inhibits stress-related increases in plasma cortisol concentration in salmon (Portz 2007). Blood will be collected from the severed caudal peduncle in 40-µl, heparinized microhematocrit capillary tubes. Blood samples from the treatment groups under the two holding conditions will be collected at 0- and 24-h posttreatment. Weights $(\pm 0.01 \text{ g})$ and measurements (TL, $\pm 1 \text{ mm}$) of each fish using an electronic balance and fish measuring board will be recorded. Collected blood will be immediately centrifuged using a microhematocrit centrifuge (Clay-Adams Autocrit Ultra3) for 4 min at $12,000 \times g$ to separate the plasma from the packed cells (Becton Dickinson Diagnostics, Sparks, Maryland). Hematocrit (packed cell volume) will be measured shortly after collection. Plasma obtained with from each fish will be transferred into plastic cryogenic freezing vials and stored in a -80°C freezer for later analyses of plasma cortisol, lactate, and glucose. Plasma cortisol concentrations will be measured using a modified enzyme immunoassay (ELISA) at the University of California, Davis Endocrinology Lab, and plasma lactate and glucose will be measured with a polarographic analyzer (YSI 2700 Select, Yellow Springs Inc., Yellow Springs, Ohio) in the Fisheries and Wildlife Group's Fish Physiology Lab. Larval delta smelt will be too small to measure plasma constituents; however other methods are being investigated.

External Tissue Damage

Scale loss and external tissue damage will be determined in the control and the treatment group immediately post-treatment using fluorescein (AK-Fluor®, Akorn, Inc., Decatur, Illinois). Fluorescein is a nontoxic fluorescent dye that can be used to rapidly and easily detect scale loss and tissue lesions and ulcers by binding to breaks or tears in the epithelial barrier of soft tissue. Fish will be euthanized in a MS-222 bath (200 mg/L) and transferred to a solution of 0.20 mg fluorescein/1ml water for 6 min and then rinsed in two separate clean water baths for 3 min each. Fish were immediately examined for skin damage under an ultraviolet light (Model UVGL-58, Mineralight, Upland, California). Photographs will be taken in complete darkness under ultraviolet light using a Nikon D-100 digital camera. Severity of tissue damage will be categorized, external bacterial infections will be diagnosed, and total damaged area will be quantified. Weights (± 0.01 g) and measurements (TL, ± 1 mm) of each fish using an electronic balance and fish measuring board will be recorded.

96-Hour Survival Monitoring

Survival will be determined over a 96-h holding period in 190-L tanks with the tanks examined daily for mortalities. Mortalities are removed daily so water quality is

not degradated. After 96 h, surviving fish will be counted, weighed (± 0.01 g), and measured (TL, ± 1 mm) using an electronic balance and fish measuring board.

Data Analyses

Statistical analyses will be performed using Sigmastat 3.0 (Jandel Scientific, San Rafael, California) software package. Differences between the treatments and control will be tested using a either a t-test or analysis of variance (ANOVA; Zar 1984, Steel *et al.* 1997). The Tukey's test will be used for all pair-wise multiple comparisons for parametric data. The Shapiro-Wilk's test for normality and the Levene's test for homogeneity of variances will be used to determine ANOVA assumptions. Data that do not meet the ANOVA assumptions and are unable to be power or log transformed will be compared with a Kruskal-Wallis non-parametric ANOVA on ranks with the Dunn's test for pairwise multiple comparisons (Zar 1984, Steel *et al.* 1997). Differences will be considered significant at P <0.05.

Coordination and Collaboration

This research will be a collaborative effort between Fisheries and Wildlife Research Group staff and TFCF biologists. Research will be coordinated directly with the Tracy Technical Advisory Team, Tracy Fish Facility Improvement Program manager and the Tracy Fish Collection staff. Opportunities for participation of state and federal resource agencies in research-related discussions at the Tracy Technical Review Team and Central Valley Fish Facilities Review Team meetings will be offered. A Tracy Series Technical Bulletin will be made available in the summer 2011.

Endangered Species Concerns

This study will not involve the use of wild endangered or threatened species. Threadfin shad (*Dorosoma petenense*) will be obtained from a commercial fish farm and larval delta smelt (*Hypomesus transpacificus*) from University of California Davis' Fish Conservation and Culture Lab in Byron, California). The laboratory evaluation of haulout bucket releases on fish damage and survival will not impact listed species.

Dissemination of Results (Deliverables and Outcomes)

The primary deliverable will be a Tracy Technical Bulletin. Technical updates may also be provided to the Tracy Technical Advisory Team. Additionally, information gained on the successes and limitations of the fish collection and salvage process will help guide future improvements in the fish collection, holding, and transport process.

Literature Cited

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